Final Abstract Number: 41.002
Session: Infectious Disease Surveillance
Date: Thursday, June 14, 2012
Time: 12:45-14:15
Room: Poster & Exhibition Area

Development of lateral flow test for the fast identification of *Vibrio Cholerae* O1

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Background: The development of simple, fast and specific methods for identification of pathogenic microbes is necessary for epidemiology control of infectious diseases. The objective of this study was to develop the fast test for user-friendly and field application in routine diagnosis of *V. cholera* ofserogroup *O1*. Among various diagnostic tools the immunochromatographic lateral flow assay (LF) it seems to be mostly perspective for fast identification of pathogens.

Methods: Use lipopolysaccharide (purified from strain *V.cholerae 569B O1*) as multi-epitopes surface antigen of *V. cholerae O1* provided getting the monoclonal antibodies (Ab). As results 21 hybridomas were generated, one pair of hibridomas has been selected and used for *V. cholerae O1* LF-test formulation. The first antibody (Ab 1) was immobilized on a membrane (test line). The second (detector) antibody (Ab 2) conjugated with colloidal gold ('Gold-Ab'), was applied to a conjugate pad.

The requirements to Ab were:

Specificity and sensitivity sufficient for application in LF format;
Preservation of the activity of Ab1 after immobilization on a surface;
Preservation of the activity of Ab2 after labeling with gold particles;

Results: The test was tried with reference collection of various *Vibrio spp*. and *sg*. It has been demonstrated that created test is suitable for identification *V. cholerae O1* after enrichment step in various media including alkaline peptone water and thiosulfate citrate bile salts sucrose agar. The test had 100% specificity and 100% sensitivity with the limit of detection – 107cfu/ml. Test had not cross-reaction with *V.cholerae O139, V.cholerae non O1/139, V.parahaemolyticus, V.mimicus, V.vulnificus*) andvarious microbes belonging to others genera (*Enterobacteriaceae, Commomonas, Aeromonas, Plesiomonas*).

Conclusion: Immunochromatographic test for identification *V. cholerae O1* with high specificity and sensitivity was developed. A new test can be used for epidemiology surveillance of cholera on identification step after enrichment of clinical or environmental samples.

http://dx.doi.org/10.1016/j.ijid.2012.05.278

Final Abstract Number: 41.003
Session: Infectious Disease Surveillance
Date: Thursday, June 14, 2012
Time: 12:45-14:15
Room: Poster & Exhibition Area

Factors related to the occurence of poliomyelitis in Nepal

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Background: In Nepal, a total of 3,116 acute flaccid paralysis (AFP) and 122 polio cases were reported between 1998 and 2009 through AFP surveillance network.

Methods: A cross sectional study was conducted to explore the factors associated with poliomyelitis cases in Nepal.

Results: It is found that the mean age of AFP cases was 5.3 years (SD = 3.9 years), while the mean age of polio cases was 4.6 years (SD = 4.0 years). Majority of polio cases were in the age group between 0.5 to 4.9 years with male preponderance. Among the polio cases, the proportion of Dalit and religious minorities were 29.7% and 13.1% respectively. Majority of polio cases were from Central development and Terai region. The proportion of polio cases was highest in the winter season (28.7%). The proportions of "zero" dose of Oral Polio Vaccine (OPV) among polio cases during routine immunization (RI), supplementary immunization activities (SIAs) and during both RI and SIAs were 29.1%, 32.3% and 14.3% respectively. There were significant associations between age ($p=0.018$), fever ($p=0.001$) and "hot case" ($p<0.001$) and the occurrence of poliomyelitis. Similarly, there were strong associations between number of OPV doses taken through RI ($p<0.001$), SIAs ($p<0.001$) and total OPV doses received though both RI and SIAs ($p<0.001$), and the occurrence of poliomyelitis.

Conclusion: Based on these findings, it is important to conduct high quality SIAs and strengthen RI to eradicate poliomyelitis from Nepal. The AFP surveillance network needs to be prioritized and strengthened in high risk areas like Central developmental and Terai region and among Dalit communities. There is need for increased awareness about "zero" dose children and "hot cases" among doctors, nurses, public health professionals working at the field level, female community health volunteers (FCHV) level, mother's group, social workers and teachers.

http://dx.doi.org/10.1016/j.ijid.2012.05.279